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Two Cases of Chronic Graft-Versus-Host Disease With Elevated Levels of Soluble Fas Ligand in Serum

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It has recently been shown that the Fas–Fas ligand (FasL) system may be one of the pathogeneses for acute graft-versus-host disease (GVHD), and it has been reported that serum soluble Fas ligand (sFasL) increases with the presence of acute GVHD. However, there is no report on a correlation between the Fas–FasL system and chronic GVHD. We present two cases of chronic GVHD with elevated levels of serum sFasL. Its level in each case was high at the onset of chronic GVHD, but it decreased with steroid therapy. Liver dysfunction also improved as the level of serum sFasL decreased. It appears in these cases that the Fas–FasL system was related to the pathogenesis of liver damage. Am. J. Hematol. 64:133–136, 2000. © 2000 Wiley-Liss, Inc.

Key words: soluble Fas ligand; chronic GVHD; liver dysfunction

INTRODUCTION

Chronic GVHD is one of the major complications that occurs at a late stage after an allogeneic bone marrow transplantation (allo-BMT) is performed. Damage is common in various tissues, including skin, liver, gut, salivary grand, lachrymal grand, lung, and so on. Not only does the patient's quality of life decrease with chronic GVHD, but this disease is also fatal in some cases. Though the pathogenesis of chronic GVHD has yet to be fully clarified, it is understood that self-reacting cytotoxic T cells attack target tissue by producing cytokines [1].

The FasL–FasL system has been implicated in one of the major cytotoxic pathways of the cytotoxic T cells. When FasL combines with Fas, an apoptotic cell death signal is transmitted to the Fas-bearing cells. Fas (Apo-1, CD95) is a member of the tumor necrosis factor (TNF) receptor family and is expressed in various tissues. FasL is a member of the TNF family and is mainly expressed on the activated T cells [2]. FasL expressed on T cells sheds a soluble form (sFasL) into the serum. Although the characteristics of sFasL is not clearly understood, it is reported that the level of serum sFasL is elevated in patients with large granular lymphocytic leukemia, natural killer cell lymphoma, and acute GVHD [3–5]. We report here on two patients with chronic GVHD whose serum sFasL levels were high at the onset of chronic GVHD, resulting in liver damage. The sFasL levels then decreased as the liver condition improved.

CASE REPORT

Case 1

The first patient is a 16-year-old male. He was admitted to our hospital on 16 January 1996 because of severe aplastic anemia. He received two types of therapies: immunosuppressive (cyclosporine) and cytokine (G-CSF). No hematological improvement resulted, however. He then underwent allogeneic bone marrow transplantation (allo-BMT) from his HLA identical sister on 18 April 1997 (day 0). Conditioning included cyclophosphamide

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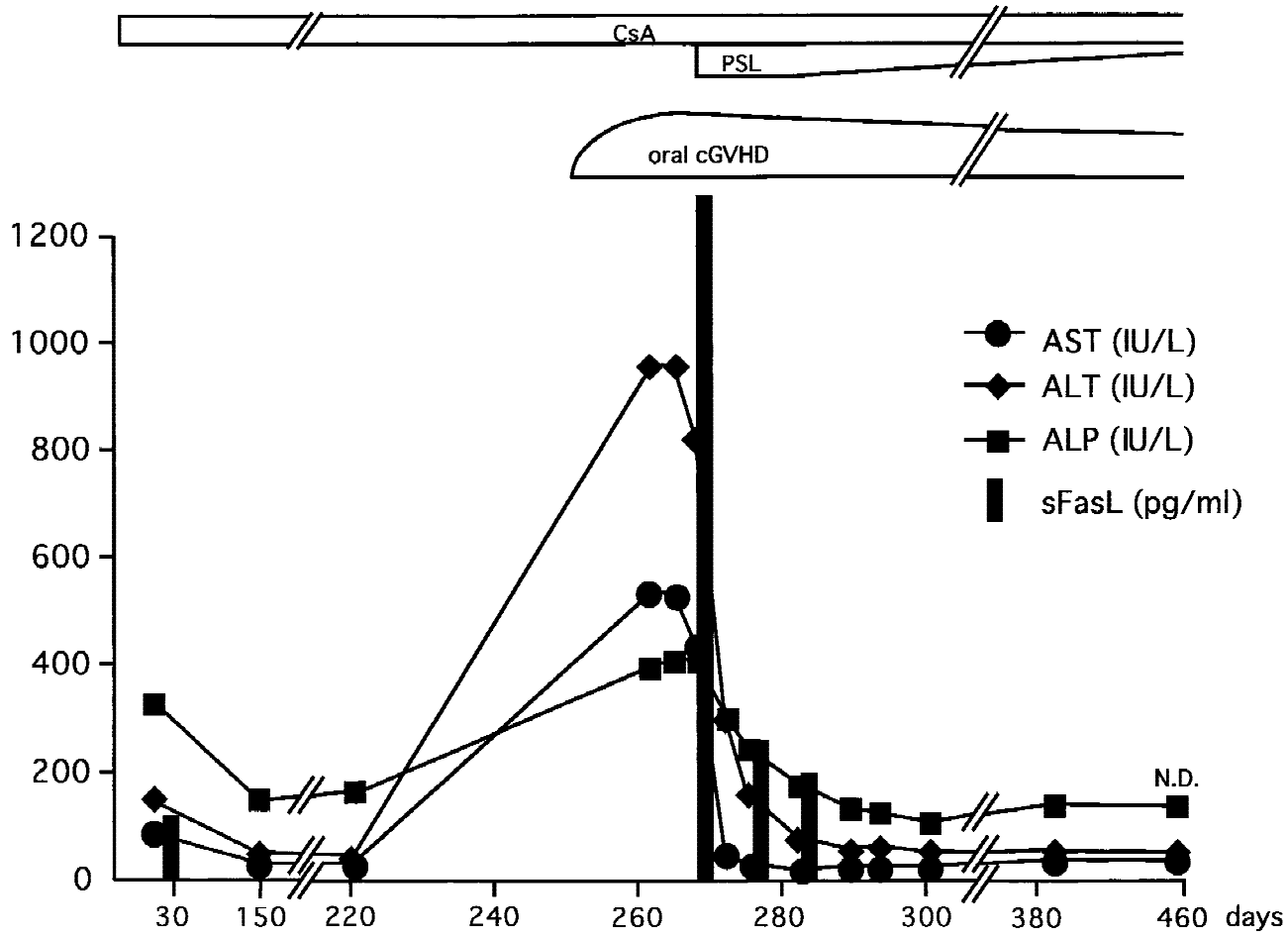

Fig. 1. Clinical course of Case 1. CsA, cyclosporine; PSL, prednisolone; cGVHD, chronic GVHD; AST, asparate aminotransferase; ALT, alanine aminotransferase; ALP, alkaline phosphatase; N.D., aFasL was not detected.

(CY) and total lymphoid irradiation. Cyclosporine (CsA) from day −1 and short-term methotrexate (MTX) were used to prevent GVHD. Although liver dysfunction was evident during the period when acute GVHD tends to occur (day 20 to day 60), we did not diagnose him as such because his liver dysfunction did not satisfy the criteria and no other symptoms of acute GVHD were present. We concluded that this liver dysfunction was a result of treatment-related toxicity and/or other drugs that were dispensed during this period. His haematopoiesis was completely restored with no occurrence of acute GVHD. He was subsequently discharged (day 123) and was followed up as an outpatient, but in December 1997 (about day 250) stomatic pain and lichen planus-like lesions of the buccal and labial mucosa appeared. A biochemical examination revealed elevated levels of liver enzymes and bilirubin, so biopsies were performed on the labial mucosa (including salivary glands) and liver. Histopathological changes were compatible with chronic GVHD, and we diagnosed him with de novo type extensive chronic GVHD. As he continued to take CsA, prednisolone (1 mg/kg) was added to suppress his immune system (day 269), and, although improvement of his stomatic condition was delayed, the liver dysfunction rapidly improved (Fig. 1).

The level of serum sFasL was chronologically measured by enzyme-linked immunosorbent assay (sFas Ligand ELISA Kit; Medical & Biological Laboratories Co., Ltd.) (Fig. 1). The sFasL levels of normal volunteers were less than 100 pg/mL in 39 of 40 samples. Only one of them showed 112 pg/mL. When acute GVHD occurs, this patient was suffering from nonspecific liver dysfunction. At that time, the level of sFasL was not elevated (day 28: 128 pg/mL), but when chronic GVHD did occur, accompanied with liver lesions, the level of sFasL did elevate (day 269: 1,240 pg/mL). As his liver condition improved by way of steroid therapy, the level of sFasL decreased dramatically. With the liver GVHD improved and the oral GVHD unchanged, sFasL could not be detected in his serum. (day 446).

He is currently 27 months post allo-BMT with a slight case of oral chronic GVHD.

Case 2

The second patient is a 49-year-old female whose diagnosis was acute myelogenous leukemia (AML) M4

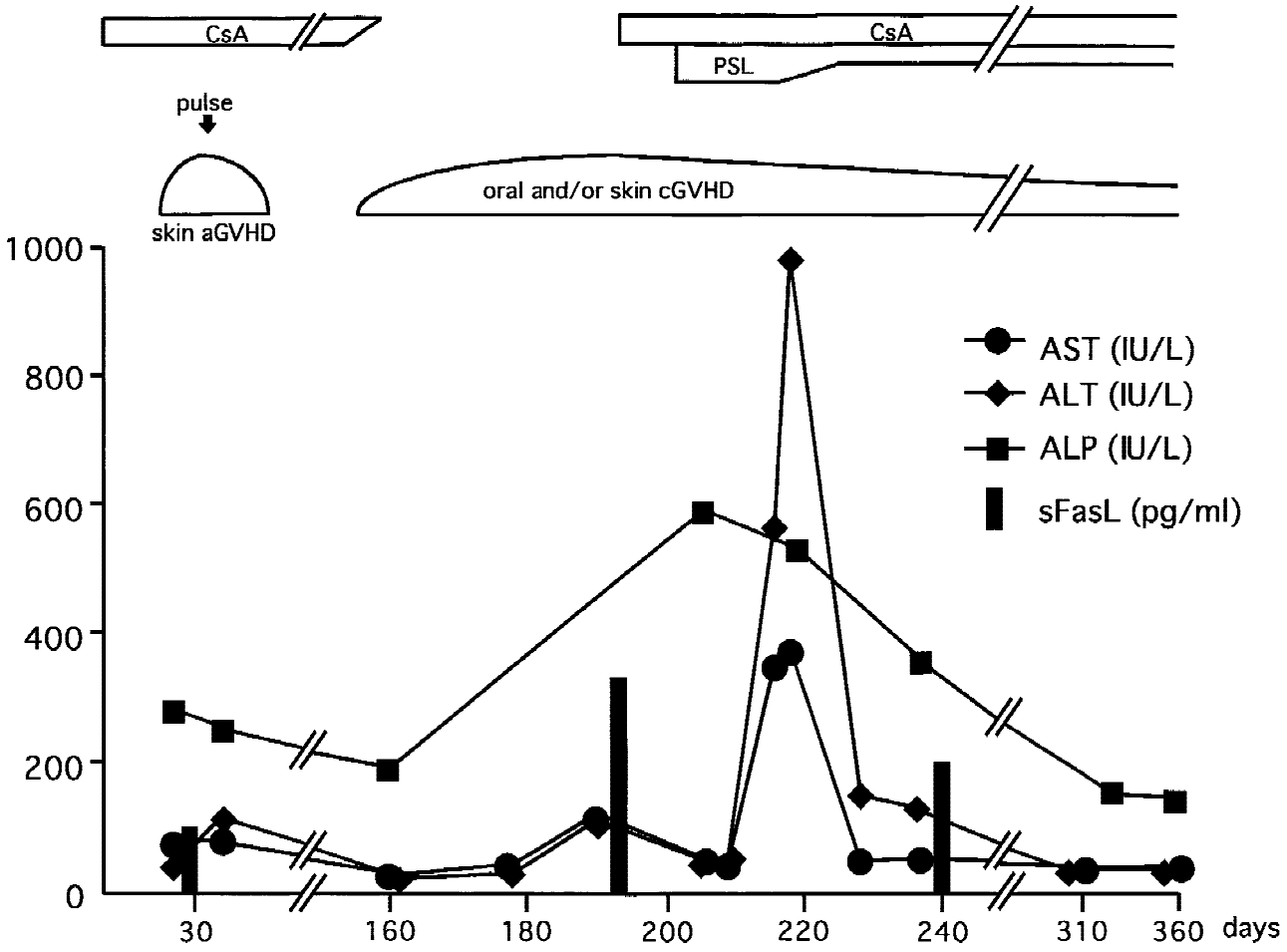

Fig. 2. Clinical course of Case 2. Pulse, steroid pulse therapy; aGVHD, acute GVHD; other abbreviations as in Figure 1.

with eosinophilia. She was admitted to our hospital because of a relapse in the central nervous system on 25 December 1996. A bone marrow examination showed no evidence of a bone marrow relapse. A whole brain irradiation was performed. She did, however, later suffer a bone marrow relapse, and a hematological complete remission (CR) was achieved with intensive chemotherapy. On 27 August 1997 (day 0), she underwent an allo-BMT from her sister whose HLA was a one-locus mismatch (B locus). Conditioning included total body irradiation and CY. CsA from day −1 and short-term MTX were used to prevent GVHD. Her hematopoiesis was completely restored, but an onset of acute GVHD limited to the skin (Stage 3, Grade 2) occurred (day 20). This was controlled with steroid pulse therapy (day 33). Lichen planus-like lesions appeared on the oral mucosa in December 1997 (about day 120) and on the skin in February 1998 (about day 160). A microscopic examination of these lesions was compatible with chronic GVHD. The skin lesions spread over more than 50% of her body surface. We diagnosed her as having a quiescent type of extensive chronic GVHD. As she continued to take CsA, prednisolone (1 mg/kg) was added on 19 March 1998 (day 204) to suppress her immune system. The level of liver enzymes began to elevate on 27 March 1998 (day 214), thus prompting us to perform a liver biopsy. Histopathological changes were compatible with chronic GVHD. Immunosuppressive therapy with CsA and prednisolone was continued, and her liver damage and skin lesions gradually improved (Fig. 2).

The level of serum sFasL was measured chronologically (Fig. 2). When this patient suffered with acute GVHD that was limited to the skin (Stage 3, Grade 2), the level of sFasL did not elevate (day 26: 100 pg/mL). When the chronic GVHD skin lesions spread, the level of sFasL did, however, elevate (day 193: 320 pg/mL), and, although there was no evidence of liver dysfunction at that time, it did appear 3 weeks later. The levels of sFasL and liver enzymes decreased when CsA and prednisolone were administered as an immunosuppressive therapy.

She is currently 23 months post allo-BMT with chronic GVHD of the skin.

DISCUSSION

FasL is a type II membrane protein that belongs to the TNF family. It is predominantly expressed on the acti-

vated T cells. When FasL combines with Fas, an apoptotic signal is transmitted to the Fas-bearing cell and apoptotic cell death is induced [2]. Like the TNF-α, about 75% of FasL is processed by matrix metalloproteinase and about 57% of it is released into the serum as sFasL [6]. Soluble TNF-α appears to be primarily responsible for the deleterious physiological responses such as cachexia and endotoxin shock, though both membrane-bound and soluble TNF-α have biological activities [7]. Regarding sFasL, it has not been shown that it induces tissue injury by apoptosis. Some reports say that sFasL induces tissue injury [8,9], but one other report says that it has a less apoptotic activity when compared with membrane-bound FasL [10]. In any case, as sFasL originates from membrane-bound FasL, the level of serum sFasL must therefore reflect the activity in the Fas–FasL system. With respect to hematological disorders, it has been reported that the levels of serum sFasL were high in the cases of large granular lymphocytic leukemia, natural killer cell lymphoma, and acute GVHD [3–5]. This paper documents our observations of two chronic GVHD patients whose serum sFasL was high. In their clinical course, the level of sFasL decreased in proportion with the improvement of liver damage. There is a different pathogenesis between acute and chronic GVHD, for example, acute GVHD occurs as a result of transplanted mature T cells attacking the recipient's organs. Chronic GVHD occurs when T cells fail to obtain and/or to maintain self-tolerance during T cell differentiation from its progenitors. Although the number of cases is very small and we have not evaluated other cases with or without cGVHD, our experience in these two cases suggests that the Fas–Fas system is also closely associated with the pathogenesis of chronic GVHD.

Both patients suffered from chronic GVHD, including liver lesions. Case 1 had the de novo type, and Case 2 had the quiescent type. Case 2 had acute GVHD that was limited to the skin (Stage 3, Grade 2) and successfully treated with steroid pulse therapy. She later developed chronic GVHD, including liver lesions. It is very interesting to note that the level of sFasL was not elevated at the onset of acute GVHD but was elevated at the onset of chronic GVHD. Although Fas is expressed in various tissues, including liver, skin, and gut, the Fas–FasL system mainly concerns itself with the pathogenesis of live injury associated with acute GVHD in the murine model [11,12]. In another study we conducted, acute GVHD patients who suffered from liver GVHD were prone to high levels of serum sFasL (date is not established). GVHD must therefore occur as a result of complex pathogenesis, and the Fas–FasL system also plays an important role, especially in liver GVHD. Thus control of the Fas–FasL system, such as the administration of anti-FasL antibody, should be considered as a new approach in the treatment of refractory chronic GVHD.

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